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REVIEW ARTICLE

Biofiltration and disinfection codetermine the bacterial antibiotic resistome in drinking water: A review and meta-analysis

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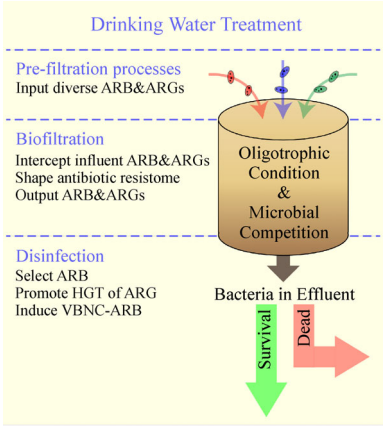
HIGHLIGHTS

- Published data was used to analyze the fate of ARGs in water treatment.
- Biomass removal leads to the reduction in absolute abundance of ARGs.
- Mechanism that filter biofilm maintain ARB/ARGs was summarized.
- Potential BAR risks caused by biofiltration and chlorination were proposed.

GRAPHIC ABSTRACT



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ABSTRACT

The bacterial antibiotic resistome (BAR) is one of the most serious contemporary medical challenges. The BAR problem in drinking water is receiving growing attention. In this study, we focused on the distribution, changes, and health risks of the BAR throughout the drinking water treatment system. We extracted the antibiotic resistance gene (ARG) data from recent publications and analyzed ARG profiles based on diversity, absolute abundance, and relative abundance. The absolute abundance of ARG was found to decrease with water treatment processes and was positively correlated with the abundance of 16S rRNA ($r^2 = 0.963$, $p < 0.001$), indicating that the reduction of ARG concentration was accompanied by decreasing biomass. Among treatment processes, biofiltration and chlorination were discovered to play important roles in shaping the bacterial antibiotic resistome. Chlorination exhibited positive effects in controlling the diversity of ARG, while biofiltration, especially granular activated carbon filtration, increased the diversity of ARG. Both biofiltration and chlorination altered the structure of the resistome by affecting relative ARG abundance. In addition, we analyzed the mechanism behind the impact of biofiltration and chlorination on the bacterial antibiotic resistome. By intercepting influent ARG-carrying bacteria, biofilters can enrich various ARGs and maintain ARGs in biofilm. Chlorination further selects bacteria co-resistant to chlorine and antibiotics. Finally, we proposed the BAR health risks caused by biofiltration and chlorination in water treatment. To reduce potential BAR risk in drinking water, membrane filtration technology and water boiling are recommended at the point of use.

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1 Introduction

The extensive use of antibiotics has resulted in a worldwide BAR problem, which poses a serious threat to human health. In recent years, ARB as well as ARGs have been

defined as emerging contaminants. More studies are focusing on environmental ARB and ARGs, among which BAR problems in drinking water are particularly worthy of attention due to their direct impact on people's daily lives (Farkas et al., 2013; Narciso-da-Rocha et al., 2013). For example, the detection of the NDM-1 gene in tap water in New Delhi, India in 2010 (Walsh et al., 2011) and the occurrence of *Escherichia coli* carrying the extended-spectrum β-lactamase (ESBL) gene in tap water in France in 2016 (Madec et al., 2016) have sounded the alarm for the biosafety of drinking water.

In drinking water treatment plants (DWTPs), biofiltration, including sand filtration and carbon filtration, is often a direct source of disinfection pressure due to ubiquitous microbial leakage problems. Recent studies have found that ARB and ARGs persist in the biofilms of drinking water biofilters (Tawfik El-Zanfaly et al., 1998;Bai et al., 2015) and may leak along with the filter effluent. Moreover, microbial leakage from the biofilter determines the bacterial community structure in the drinking water distribution system (DWDS, Pinto et al., 2012), and it may further shape the antibiotic resistome in pipeline biofilm (Jia et al., 2015). These studies indicate that biofilters pose significant biosafety risks and may play a key role in the BAR problem in drinking water.

Before entering the distribution pipeline system and then the point of use (POU), most drinking water is disinfected in drinking water treatment plants (DWTPs). Disinfection will kill/inactivate most bacteria, and removal efficiencies of several logs can usually be reached using chlorination/chloramination, ozonation, ultraviolet (UV) treatment, and other methods. Laboratory experiments have revealed, however, that intracellular ARGs present lower damage rates in disinfection compared to the inactivation rates of bacterial cells (McKinney and Pruden, 2012; Yoon et al., 2017), and disinfection techniques such as chlorination may actually result in the enrichment of ARGs in disinfected water (Murray et al., 1984). In recent years, in-depth research on disinfection has found that chlorine and UV light cannot kill bacteria completely, but induce bacterial cells into a viable but nonculturable (VBNC) state (Zhang et al., 2015; Chen et al., 2018). The above studies indicate that disinfected water from DWTPs may not only serve as the ARG pool for pipeline biofilm, but they also discharge active ARB into tap water.

Based on the above, biofiltration is, in fact, both a sink and a source of ARB and ARGs. Disinfection can greatly affect the overall microbial community and ARB/ARG diversity. Until now, however, no comprehensive review has been conducted on the distribution and fate of ARGs throughout the drinking water treatment process. In this review, we first collated the occurrence and distribution of ARB/ARGs during water treatment and then assessed the roles of biofiltration and chlorination in shaping the bacterial antibiotic resistome. Our objectives were to: (1) profile the bacterial antibiotic resistome in different drinking water treatment processes by meta-analyzing the published data on absolute abundance, relative abundance, and diversity of ARGs; (2) identify the contributions of key biofiltration and disinfection treatment units on the bacterial antibiotic resistome and their influence on ARB and ARGs; and (3) assess the microbial health risk of the BAR caused by biofiltration and disinfection of drinking water.

2 Data collection and meta-analysis

To survey the BAR problem in water treatment, we used Google Scholar to retrieve publications. The search terms included "antibiotic resistant bacteria" AND/OR "antibiotic resistance genes" AND/OR "drinking water treatment" AND/OR "potable water treatment." The database was searched for studies published from 1990 to 2018. A total of 1,890 publications were found. Publications were then checked individually to eliminate any duplicates or irrelevant articles. Relevant articles focusing on screened strains in drinking water treatment were also discarded considering the difference of BAR profile between the screened colonies and the whole bacterial community. Finally, 17 publications reporting BAR in drinking water treatment were selected (Table S1). The data in relevant publications were extracted for further analysis using Plot Digitizer software (plotdigitizer.sourceforge.net).

Technically, ARG detection is usually based on different molecular methods: quantitative PCR (qPCR), high-throughput quantitative PCR (HT-qPCR), and high-throughput sequencing (HTS). qPCR is used to quantify the content of certain ARGs, but has a low flux. Therefore, it is rarely used for broad-spectrum scans of ARGs at the resistome level. HT-qPCR improves the throughput and reduces the experimental cost compared with qPCR. By designing appropriate primers, HT-qPCR is capable of detecting the ARGs of the entire antibiotic resistome in a single experiment. The combined use of HTS and bioinformatics analysis can also achieve the goal of scanning the bacterial antibiotic resistome. In this study, the data obtained using qPCR were used to profile ARGs at the genetic level, while the data obtained using HT-qPCR and HTS were utilized to profile ARGs at the resistome level.

3 Distribution of ARGs in DWTPs

To identify the ARG profiles in different treatment processes, data were collected and analyzed from 3 perspectives: the diversity of ARGs (i.e., the richness of ARGs, the Shannon index of ARGs, and the Simpson index of ARGs), the absolute abundance of ARGs (gene copies of ARGs per unit water volume), and the relative abundance of ARGs (proportion of normalized ARG copies to 16S rRNA gene copies).

3.1 Diversity of ARG

Data of ARG diversity from one study using HTS in conventional water treatment processes (Jia et al., 2015) and two studies using HT-qPCR in advanced water treatment processes (Xu et al., 2016; Zheng et al., 2018) were extracted (Fig. 1). The results of HTS revealed that sedimentation and chlorination can effectively reduce ARG diversity, while sand filtration leads to an increase in ARG diversity. However, in other study utilizing HT-qPCR, result revealed that sedimentation and sand filtration exhibit little effect on the diversity of ARG, while the richness of ARG decreased from 70 to 67 after sedimentation and from 67 to 65 after sand filtration. In the two studies using HT-qPCR, no significant difference ($p>0.05$) was found between ARG richness in treatment processes, and the variation trend of ARG richness in water treatment process was consistent, and there was no significant difference ($p>0.05$) between the two groups of data. It is worth noting that GAC filtration resulted in a significant increase in ARG diversity (paired-*t*-test, $p<0.05$), and the results of 2 publications showed that ARG richness increased from 76 to 150 and from 80 to 160 after GAC filtration.

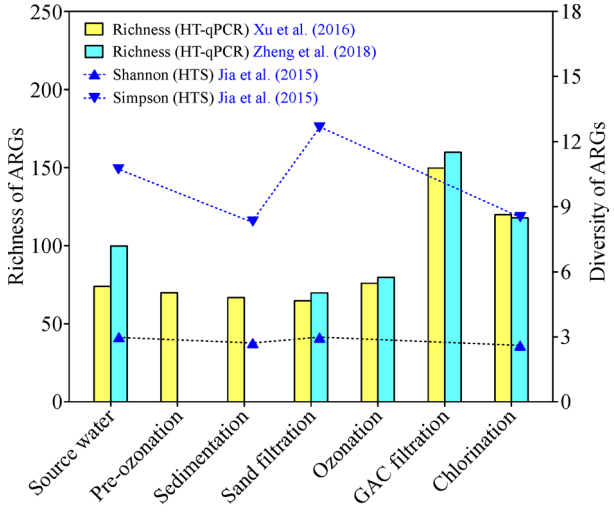


Fig. 1 The diversity of ARGs in different DWTPs.

3.2 Absolute abundance of ARG

Data based on qPCR and HT-qPCR methods were extracted from four studies (Xu et al., 2016; Zhang et al., 2016; Su et al., 2018; Zheng et al., 2018) to indicate the change of absolute ARG abundance in drinking water treatment. From source water to disinfected water, ARGs decreased in absolute abundance along the water treatment processes (Figs. 2(a) and 3(a)), similar results were also found in other studies (Bergeron et al., 2015; Bergeron et al., 2017; Hu et al., 2018; Lu et al., 2018). For all of the detected ARGs, sand filtration contributed the highest proportion of ARG removal (97%), followed by sedimentation (70.9%), chlorination (68.8%), and ozonation + GAC (58.3%) (Fig. 2(b)). At the resistome level, three DWTPs exhibited significant different structure (ANOVA, $p<0.05$) in the absolute abundance of ARG in source water, however, the attenuation trend of ARGs was consistent along treatment process among studies. In addition, the ARG abundance was found to be correlated with the 16S rRNA gene abundance significantly and positively ($p<0.001$; Fig. 3(b)), suggesting that ARG removal at the resistome level was combined with the reduction of bacterial biomass in water treatment. It is well known that bacteria can be effectively removed by sedimentation (Bustamante et al., 2001), sand filtration (Bomo et al., 2004; D'Alessio et al., 2015), and chlorination (Ma and Bibby, 2017), while microbial leakage leads to the high bacterial cell concentrations found in the effluent of GAC filters (Korotta-Gamage and Sathasivan, 2017). As a result, it is reasonable that the processes of sedimentation, sand filtration, and chlorination can remove ARGs effectively, while carbon filtration cannot.

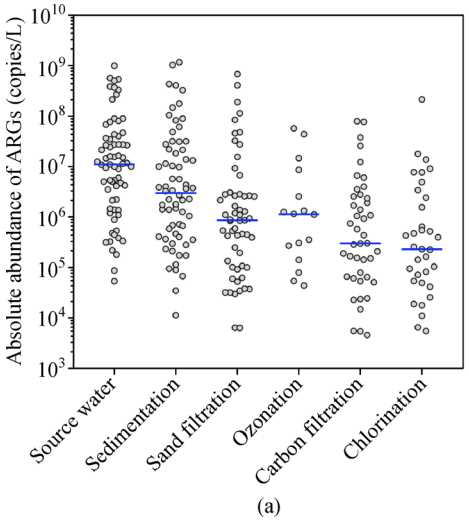
3.3 Relative abundance of ARGs

The qPCR results suggested that 50% of the detected ARGs increased in relative abundance with treatment processes, while the other 50% decreased (Fig. 4). It is worth noting that the enriched ARGs exhibited a significant increase in amplification after either GAC filtration or chlorination. At the resistome level, the HT-qPCR results suggested that coagulation + sedimentation, sand filtration, and ozonation demonstrated little influence on the relative abundance of ARG, while GAC filtration and chlorination not only increased ARG abundance significantly (paired-*t*-test, $p<0.05$) but also changed the structure of the antibiotic resistome (Fig. 5). Similar results showing that sand filtration slightly changes the relative abundance of ARG at the resistome level were also found by other studies using HTS (Fig. 6, drinking water treatment; Christgen et al., 2015; Jia et al., 2015).

4 Impact of biofiltration and disinfection on the occurrence of ARGs and ARB

4.1 Impact of biofiltration on the occurrence of ARGs and ARB

The impacts of sand filtration on ARG diversity vary among different studies (Fig. 1). The results of qPCR suggest that sand filtration effectively reduces the absolute abundance of ARGs (Fig. 2), with 97% of the detected ARGs decreasing in absolute abundance, a higher proportion than either sedimentation (70.9%), ozonation + GAC (58.3%), or chlorination (68.8%). At the resistome level,

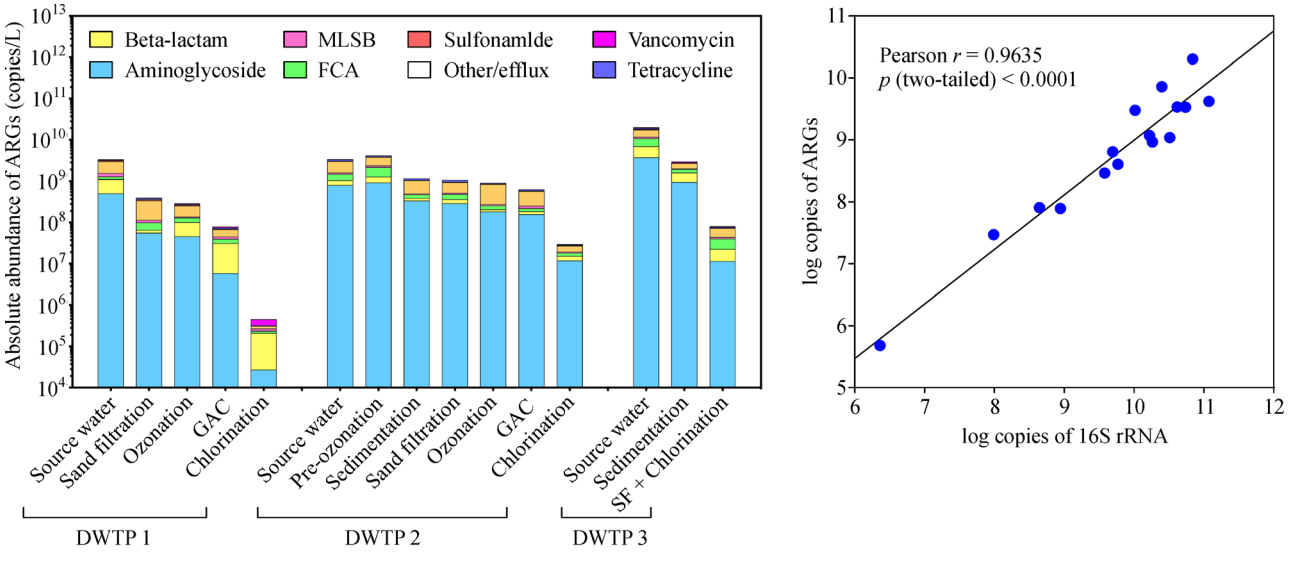


(a)

ARG removal	Sedimentation	Sand filtration	Ozonation + GAC	Chlorination
⩾2 log	1.4%	10.1%	10.4%	18.8%
1~2 log	15.9%	33.3%	2.1%	14.6%
0~1 log	53.6%	53.6%	45.8%	35.4%
−1~0 log	29.0%	1.4%	29.2%	20.8%
−2~−1 log	0.0%	1.4%	4.2%	6.3%
⩽−2 log	0.0%	0.0%	8.3%	4.2%
	Among 69 detected ARGs		Among 48 detected ARGs	

(b)

Fig. 2 The impact of treatment processes on the absolute abundance of ARG based on qPCR. (a) Distribution of ARG (copies/L) in different drinking water treatment processes, the detail data are listed in Table S2; (b) Log removal of ARG by different treatment processes, the detail data are listed in Table S3.



(a) (b)

Fig. 3 The impact of treatment processes on the absolute abundance of ARG based on HT-qPCR. (a) Distribution in absolute abundance of each type of ARG. MLSB represents macrolide lincosamide-streptogramin B resistance genes, FCA represents fluoroquinolone, quinolone, florfenicol, chloramphenicol, and amphenicol resistance genes; (b) Correlation between the absolute abundance of ARG and 16S rRNA, the detail data are listed in Table S4.

the relative abundance of ARGs changed slightly but the antibiotic resistome structure shifted after sand filtration (Figs. 5 and 6). As mentioned earlier in this manuscript (Section 3), ARG removal is accompanied by the reduction of biomass. In DWTPs, sand filtration possesses the efficacy to remove microorganisms (Devi et al., 2008), which may further result in the reduction of absolute ARG abundance. However, the detachment of the filter biofilm releases the bacterial cells into the filter effluent. Since the filter biofilm has a different bacterial community structure than the filter influent, the finding that the antibiotic resistome and ARG diversity changed after sand filtration is reasonable.

Based on the results of Section 3, GAC filtration exhibited weaker effects on ARG removal than sand filtration. After the ozonation + GAC treatment, either the richness or the relative abundance of ARG increased, accompanied by changes in the antibiotic resistome (Fig. 5). Ozonation could improve the biodegradability of the organic matter in the water by breaking macro-

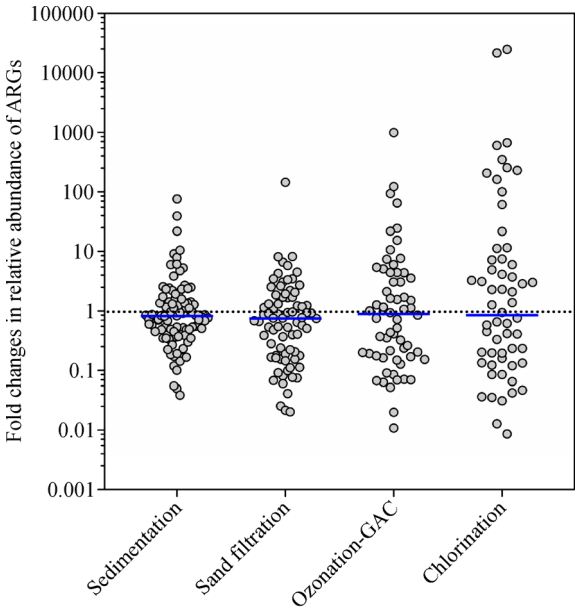


Fig. 4 Fold changes in relative abundance of ARGs after treated with different processes compared to the source water. The data are obtained by using HT-qPCR and is listed in Table S5.

molecular organic matter into lower-molecular matter (Narkis and Schneider-Rotel, 1980; Legube et al., 1981), which will be utilized by GAC filter biofilm. Various studies have shown that GAC filters harbor a large biomass of highly diverse bacteria (Weber et al., 1978; Brewer and Carmichael, 1979; Li et al., 2017; Zhu et al., 2019), making it possible to maintain diverse ARGs in biofilm (Fig. 5(b)). In addition, due to the prevalence of microbial leakage in carbon filtration processes (Han et al., 2013; Liu et al., 2016(b)), the shedding biofilm and the embedded ARB/ARGs are further input into downstream processes, thus posing health risks.

4.2 Impact of disinfection on the occurrence of ARGs and ARB

Chlorination can significantly reduce the diversity and richness of ARGs (Fig. 3). Results of HTS indicated that the Shannon index decreased from 3.0 to 2.6 while the Simpson index decreased from 13 to 8. Results of HT-qPCR revealed that the richness decreased from 150 to 120 and from 160 to 120 in two DWTPs. According to Fig. 2(b), 68.8% of the ARGs decreased in absolute abundance after chlorination, with 18.8% of the detected ARGs decreasing by more than 2 logs, a proportion that exceeded that of sedimentation (1.4%), sand filtration (10.1%), and ozonation + GAC (10.4%). Overall, these results indicated that chlorination exhibits a positive effect in the reduction of ARG diversity and absolute abundance. However, chlorination has a limited impact on controlling the relative abundance of ARGs. Instead, significant increases in relative ARG abundance, from 0.09 to 0.2, 0.13 to 0.3, and 38 to 94.31 ppm, were found after chlorination in different studies based on HT-qPCR and HTS (Figs. 5 and 6), and qPCR results revealed that the enrichment of some types of ARGs may lead to increasing ARG abundance at the resistome level (Xi et al., 2009; Guo et al., 2014; Zhang et al., 2016; Su et al., 2018). These results further suggest that a portion of the ARGs as well as their carriers were enriched by chlorination. Previous research has proven that chlorination has a strong effect in shaping the structure of the bacterial community (Shi et al., 2013). After chlorination, the surviving bacteria are more resistant to chlorine (Murray et al., 1984) and the

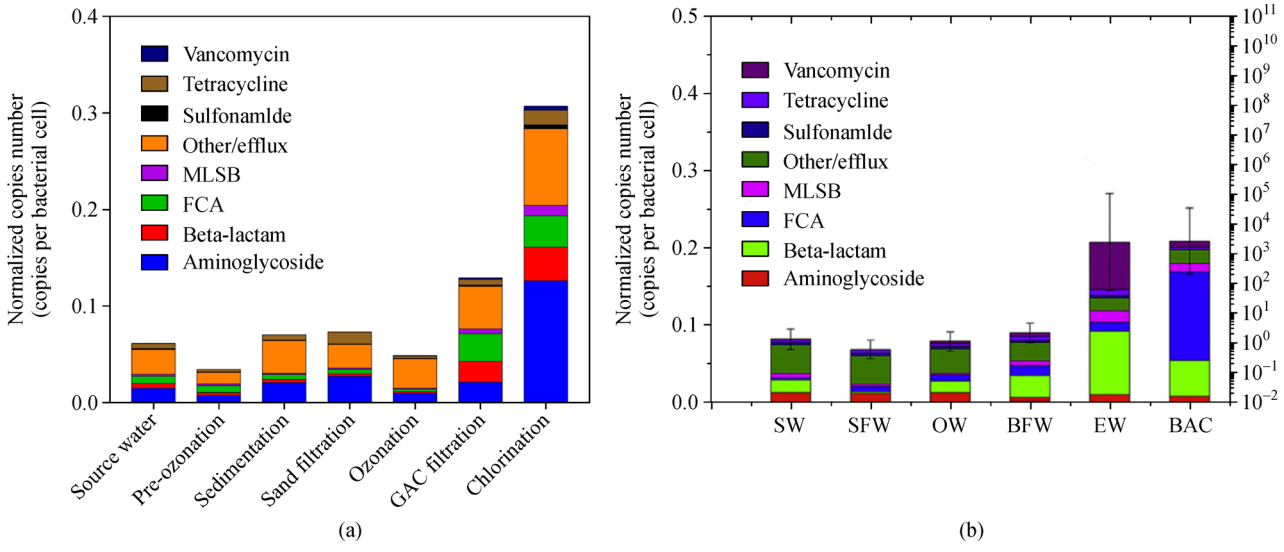


Fig. 5 The impact of treatment processes on the relative abundance of ARG at resistome level. b. "SW," "SFW," "OW," "BFW," "EW" and "BAC" represent source water, sand filtered water, ozone water, BAC filtered water, effluent and biofilm on GAC filter, respectively. Data of (a) was extracted from Xu et al. (2016); (b) was adapted from Zheng et al. (2018) with permission from author.

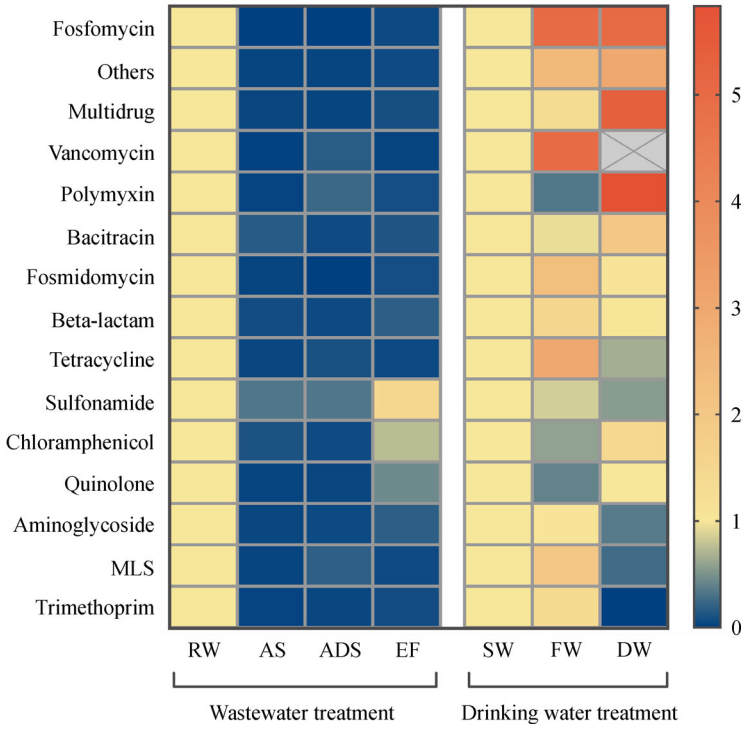


Fig. 6 Fold changes in relative abundance of ARGs after treated with different processes compared to the raw water. The data are obtained by using HTS. Data in wastewater treatment processes was extracted from Christgen et al. (2015), while data in drinking water treatment processes was extracted from Jia et al. (2015). "RW," "AS," "ADS," "EF," "SW," "FW" and "DW" represents raw wastewater, active sludge, anaerobic digestion sludge, effluent, source water, filtered water and disinfected water, respectively.

proportion of bacteria co-resistant to both chlorine and antibiotics increases (Xu et al., 2016).

5 BAR risk caused by biofiltration and disinfection

5.1 Enrichment of ARGs and ARB by biofiltration

Bacterial communities in source water are usually associated with diverse and varying structures, which may carry different kinds of ARGs. The occurrence and diversity of ARGs in surface water have been reviewed (Yang et al., 2018). In DWTPs, most source water bacteria will be trapped and removed during filtration (Bomo et al., 2004; Hijnen et al., 2010), while ARGs as well as their carriers may attach to the filter biofilm. To obtain the resources (nutrients, space) necessary for survival, the trapped bacteria will compete with the indigenous bacteria (Zhang et al., 2011). During this process, the ARG-carrying bacteria, either derived from influent or the indigenous biofilm, may persist and become further enriched in the biofilter (Li et al., 2018).

5.2 Characteristics of filter biofilm maintain ARB and ARGs

Bacterial antibiotic resistance can be recognized as the increase of the minimum inhibitory concentration value of an antibiotic caused by genetic changes in bacteria that may be attributed to either mutation or horizontal gene transfer (HGT). By contrast, bacterial antibiotic tolerance is the ability to resist antibiotics due to reversible phenotypic changes such as transient, dormant, and non-dividing states (Mah, 2012). In drinking water biofilters, both bacterial resistance and tolerance exist. In addition, biofilms composed of bacterial cells and extracellular polymeric substances (EPS) are tolerant to antibiotics as a whole and protect the bacteria within them.

5.2.1 Antibiotic tolerance in filter biofilm

Protection of the matrix. The biofilm matrix is mainly composed of proteins, extracellular polysaccharides, lipids, and DNA. One of the functions of biofilm is to physically prevent the penetration of antibiotics (Walters et al., 2003; Chiang et al., 2013). Another function is to play the role of the digestive system in the binding and consuming of antibiotics. The extent of such functions may be affected by antibiotic type, as well as the age and heterogeneity of the biofilm. Several studies have shown that biofilm polysaccharides play the active role in protecting bacterial cells from antibiotics (Bagge et al., 2004; Van Acker et al., 2014). Additionally, it is worth noting that bacterial cells within biofilm present higher tolerance to antimicrobial agents, including antibiotics

such as tobramycin and their planktonic counterparts (Hoyle and Costerton, 1991; Hill et al., 2005; Tabak et al., 2007). Such bacterial tolerance within biofilm is reported to be caused by low metabolic activity (Brown et al., 1988), the scarcity of nutrients (Bernier et al., 2013), and oxygen (Borriello et al., 2004).

5.2.2 Filter biofilm promotes mutations and HGT

Due to the stress caused by the scarcity of nutrients, oxygen, and the accumulation of extracellular metabolites, antibiotics in biofilm may increase the intracellular reactive oxygen species (ROS) (Sakai et al., 2006), which will damage DNA and may further promote mutations (Friedberg et al., 2005). It was reported that *P. aeruginosa* grown in a biofilm state showed a 105-fold increase in mutability compared to that in planktonic cells (Driffield et al., 2008). Conibear et al. found that microcolony-based growth also promoted the mutagenicity of *P. aeruginosa* cells more than 100-fold (Conibear et al., 2009). In addition, biofilm is the ideal environment for HGT, given its stable physical structure, which is conducive for cell-cell contact and the persistence of extracellular DNA (reviewed in Molin and Tolker-Nielsen, 2003 and Madsen et al., 2012). It has been reported that gene transfer either by plasmid conjugation or DNA transformation occurs frequently and effectively in natural and bioreactor/laboratory-based biofilm (Beaudoin et al., 1998; Normander et al., 1998; Geisenberger et al., 1999; Roberts et al., 1999). In drinking water biofilters, the intercepted mobile genetic elements and their resident ARGs are likely to persist or even spread in the filter biofilm.

5.3 Microenvironments improve the fitness of ARB in filter biofilm

The BAR is often associated with reduced bacterial fitness. It is speculated that the expression of ARGs consumes extra energy or engenders bacterial structural and functional modifications, and the resistant bacteria would be overcome by their susceptible counterparts as the dosage of antibiotic decreases (Levin et al., 1997). The fitness of ARB is closely related to their survival environment and is affected by various environmental factors (Andersson and Hughes, 2010). In drinking water biofilters, the factors that may affect the fitness of ARB are listed as follows.

5.3.1 Antibiotics

Antibiotics are the primary factor affecting ARB fitness. When the environmental antibiotic level is above the minimal inhibitory concentration (MIC), sensitive bacteria cannot survive while resistant bacteria will bloom with low competitive pressure. At the sub-MIC level, although it is difficult for antibiotics to select ARB directly, there is in theory a minimum selective concentration (MSC) of antibiotics for ARB to out-compete their susceptible counterparts, which are limited in growth rate in the presence of antibiotics (Baquero et al., 2008; Negri et al., 2000). Such a theory was proved by laboratory experiments demonstrating that resistant mutants of *S. typhimurium* (streptomycin) and *E. coli* (ciprofloxacin) could be enriched in batch cultures by competing with their isogenic wild type strains at the sub-MIC antibiotic level (1/4 MIC for *S. typhimurium* and 1/10 MIC for *E. coli*) (Gullberg et al., 2011; Liu et al., 2011). It is worth noting that sub-MIC selection is concentration-limited and often takes an extended period of time. Andersson et al. summarized the lowest MSC (1/230 MIC), which was found in the clinical isolates of fluoroquinolone-resistant *E. coli* (Andersson and Hughes, 2012).

5.3.2 Heavy metals

Bacterial metal resistance and antibiotic resistance can be genetically (co-resistance) and physiologically (cross-resistance) linked (Seiler and Berendonk, 2012); such linkages make it possible for heavy metals to co-select ARB (Icgen and Yilmaz, 2014). This metal-driven co-selection has been found in mercury (Hg), cadmium (Cd), copper (Cu), zinc (Zn), etc. (Summers et al., 1993; Ghosh et al., 2000; Hasman and Aarestrup, 2002; Dean et al., 2007), and is affected by the concentration of metal and species of bacteria (Nies, 2000; Akinbowale et al., 2007; Abskharon et al., 2008). It is worth noting that environmental conditions can affect the bioavailability of heavy metals by influencing their valence, further affecting the co-selection of ARB (Seiler and Berendonk, 2012). Although many studies have focused on the co-selection of ARB driven by heavy metals, to date, the MSC of such co-selection has rarely been studied systematically.

5.3.3 Organic carbon

In addition to selective pressures, typical environmental factors, such as organic carbon, can also influence the fitness of ARB by affecting the relative generation time/growth rate between ARB and their susceptible counterparts (Andersson and Hughes, 2010). Bench experiments have revealed that streptomycin-resistant strains induced by *rpsL* mutation grow faster than wild types in media with poorer carbon sources (Paulander et al., 2009). Lin et al. further found that oligotrophic conditions lead to the reduction in fitness cost of rifampin-resistant strains and facilitate the persistence of rifampin resistance (Lin et al., 2018). The above results are based on competition experiments of selected strains at the community level. A recent study on pilot sand filters found that poorer organic carbon concentrations result in greater diversity and relative abundance of ARGs in biofilm, indicating that an

oligotrophic environment facilitates the persistence of ARGs in drinking water biofilters (Wan et al., 2019).

The source water used for drinking water treatment is usually clean surface water or groundwater, which makes selective pressures such as antibiotics extremely low (ppt or ppb levels), and frequently not even detectable (Ye et al., 2007; Benotti et al., 2009; Simazaki et al., 2015; Liu et al., 2016a). Such low selective pressures make ARB selection scarcely possible during water treatment. One overlooked fact is that drinking water is typically an oligotrophic environment, and the low nutritional conditions will reduce the fitness cost of the BAR, which is conducive to the maintenance of ARB/ARGs and makes it possible for the ARB/ARGs intercepted by drinking water biofilters to persist. Heavy metals may also contribute to the persistence of the BAR in drinking water biofilters, a hypothesis supported by a previous study that detected bacteria co-resistant to copper and antibiotics in filter biofilm (Zhang et al., 2018b). However, the selective concentration of different heavy metals remains unclear in drinking water treatment.

5.4 The limitations of disinfection when faced with the BAR problem

As a post-filtration process, chlorination is subject to BAR pressure caused by the microbial leakage of biofilters. Although chlorination enhances the reduction of the absolute abundance and diversity of ARGs, this treatment still has limitations when dealing with ARB and ARGs.

5.4.1 Chlorination selects ARB

The bacterial community structure and interspecies relationships will determine the effects of chlorine disinfection, and the shifted bacterial community may subsequently result in the alteration of the antibiotic resistome (Berry et al., 2006; Wang et al., 2018). Many studies have found that chlorination exhibits a positive effect on the removal of ARB/ARGs, but ARGs as well as ARB can be enriched after chlorination (Armstrong et al., 1982; Rusin and Gerba, 2001; Huang et al., 2011). Such results are likely due to the selection of co-resistant bacteria to both antibiotics and chlorine during chlorination (Chapman, 2003; Chao et al., 2013; Khan et al., 2016; Garner et al., 2018). It has been reported that extracellular stress can facilitate the replication of bacterial intracellular plasmids (Wegrzyn and Wegrzyn, 2002). During water disinfection, the stress of extracellular chlorine might also increase the copies of ARG-carrying plasmids in the bacterial cells.

5.4.2 Low chlorine doses may promote HGT of ARGs

Guo et al. found that the frequency of conjugation in antibiotic-resistant *E. coli* strains was significantly promoted 2 to 5–fold after chlorination (Guo et al., 2015), which was likely due to the increasing pilus or pores on the bacterial cells that were treated with chlorine. As an oxidant, chlorine can damage the cell membranes of bacteria, leading to the release of ARGs. In wastewater treatment, chlorination was found to increase both the concentration and the abundance of extracellular ARGs (eARGs) (Zhang et al., 2018c; Liu et al., 2018). A previous study proved that environmental plasmid-borne eARGs could persist for a long period of time and have the potential to spread to bacterial cells via transformation (Mao et al., 2014). Overall, the above studies highlight the risk of chlorine-induced HGT of ARGs, which is likely to be present in drinking water disinfection.

5.4.3 Chlorination induces ARB into a VBNC state

Recent studies have found that disinfectants such as UV light, chlorine, and monochloramine can induce bacterial cells, including ARB cells, into a VBNC state (Turetgen, 2008; Liu et al., 2009; Zhang et al., 2015). Experiments on *E. coli* revealed that the culturable cells decreased from 10^6 to 0 per mL in chlorination at doses of 5–240 (mg × min)/L, while the viable cells remained at approximately 10^3–10^5/mL (Chen et al., 2018). Although such VBNC-state bacterial cells seldom proliferate, they retain metabolic activity, potential virulence, and cellular integrity, and exhibit greater antibiotic tolerance than their culturable counterparts (Steinert et al., 1997; Lin et al., 2017). It is worth noting that VBNC-state bacterial cells have the potential to resuscitate their proliferative ability as environmental conditions change (Steinert et al., 1997; Du et al., 2008). Such resuscitation will pose health risks when these bacterial cells arrive at the POU and are consumed by humans (Zhang et al., 2018a).

6 Conclusions: Filtration and disinfection play key roles in the drinking water BAR problem

From source water to disinfected water, ARG concentrations decrease with the reduction of biomass. Meanwhile, the profile of the bacterial antibiotic resistome changes during water treatment. In this process, biofiltration and chlorination play key roles. Biofiltration enriches the influent ARB and ARGs and maintains the BAR in biofilm, which provides ARG pools for the DWDS, while chlorination exhibits a selective effect on ARB and ARGs before entering the DWDS.

Biofiltration intercepts most influent bacteria, including ARG-carrying bacteria. The competition between the intercepted bacteria and the indigenous bacteria determines the bacterial community structure of the filter biofilm and

shapes the antibiotic resistome. The detachment of filter biofilm exports ARB and ARGs downstream. Compared to the biofilter influent, the effluent ARGs may either increase or decrease in absolute abundance, but they present differences in species, diversity, and relative abundance. Chlorination further selects ARB and ARGs and feeds them into the water distribution pipeline. The combination of biofiltration and chlorination may output ARB and ARGs that either: (1) directly reach the POU and cause a biosafety risk or (2) seed on the pipeline biofilm, thereby affecting its bacterial community structure and antibiotic resistome, and ultimately affecting tap water. To prevent the biosafety risks caused by drinking water BAR, membrane filtration such as ultrafiltration applied at the POU is recommended. Another simple method is to boil the tap water in order to inactivate the bacterial cells before drinking.

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References

Abskharon R N N, Hassan S H A, Gad El-Rab S M, Shoreit A A M (2008). Heavy metal resistant of *E. coli* isolated from wastewater sites in Assiut City, Egypt. Bulletin of Environmental Contamination and Toxicology, 81(3): 309–315

Akinbowale O L, Peng H, Grant P, Barton M D (2007). Antibiotic and heavy metal resistance in motile aeromonads and pseudomonads from rainbow trout (*Oncorhynchus mykiss*) farms in Australia. International Journal of Antimicrobial Agents, 30(2): 177–182

Andersson D I, Hughes D (2010). Antibiotic resistance and its cost: Is it possible to reverse resistance? Nature Reviews. Microbiology, 8(4): 260–271

Andersson D I, Hughes D (2012). Evolution of antibiotic resistance at non-lethal drug concentrations. Drug Resist. Updat., 15(3): 162–172

Armstrong J L, Calomiris J J, Seidler R J (1982). Selection of antibiotic-resistant standard plate count bacteria during water treatment. Applied and Environmental Microbiology, 44(2): 308–316

Bagge N, Hentzer M, Andersen J B, Ciofu O, Givskov M, Høiby N (2004). Dynamics and spatial distribution of beta-lactamase expression in *Pseudomonas aeruginosa* biofilms. Antimicrobial Agents and Chemotherapy, 48(4): 1168–1174

Bai X, Ma X, Xu F, Li J, Zhang H, Xiao X (2015). The drinking water treatment process as a potential source of affecting the bacterial antibiotic resistance. Science of the Total Environment, 533: 24–31

Baquero F, Negri M C, Morosini M I, Blazquez J (2008). The antibiotic selective process: concentration-specific amplification of low-level resistant populations. Antibiotic Resistance: Origins, Evolution, Selection and Spread, 207, 93

Beaudoin D L, Bryers J D, Cunningham A B, Peretti S W (1998). Mobilization of broad host range plasmid from Pseudomonas putida to established biofilm of *Bacillus azotoformans*. I. Experiments. Biotechnology and Bioengineering, 57(3): 272–279

Benotti M J, Trenholm R A, Vanderford B J, Holady J C, Stanford B D, Snyder S A (2009). Pharmaceuticals and endocrine disrupting compounds in U.S. drinking water. Environmental Science & Technology, 43(3): 597–603

Bergeron S, Boopathy R, Nathaniel R, Corbin A, LaFleur G (2015). Presence of antibiotic resistant bacteria and antibiotic resistance genes in raw source water and treated drinking water. International Biodeterioration & Biodegradation, 102: 370–374

Bergeron S, Raj B, Nathaniel R, Corbin A, LaFleur G (2017). Presence of antibiotic resistance genes in raw source water of a drinking water treatment plant in a rural community of USA. International Biodeterioration & Biodegradation, 124: 3–9

Bernier S P, Lebeaux D, DeFrancesco A S, Valomon A, Soubigou G, Coppée J Y, Ghigo J M, Beloin C (2013). Starvation, together with the SOS response, mediates high biofilm-specific tolerance to the fluoroquinolone ofloxacin. PLoS Genetics, 9(1): e1003144

Berry D, Xi C, Raskin L (2006). Microbial ecology of drinking water distribution systems. Current Opinion in Biotechnology, 17(3): 297–302

Bomo A M, Stevik T K, Hovi I, Hanssen J F (2004). Bacterial removal and protozoan grazing in biological sand filters. Journal of Environmental Quality, 33(3): 1041–1047

Borriello G, Werner E, Roe F, Kim A M, Ehrlich G D, Stewart P S (2004). Oxygen limitation contributes to antibiotic tolerance of *Pseudomonas aeruginosa* in biofilms. Antimicrobial Agents and Chemotherapy, 48(7): 2659–2664

Brewer W S, Carmichael W W (1979). Microbiological characterization of granular activated carbon filter systems. Journal- American Water Works Association, 71(12): 738–740

Brown M R, Allison D G, Gilbert P (1988). Resistance of bacterial biofilms to antibiotics: A growth-rate related effect? J. Antimicrob. Chemother., 22(6): 777–780

Bustamante H A, Shanker S R, Pashley R M, Karaman M E (2001). Interaction between *Cryptosporidium oocysts* and water treatment coagulants. Water Research, 35(13): 3179–3189

Chao Y, Ma L, Yang Y, Ju F, Zhang X X, Wu W M, Zhang T (2013). Metagenomic analysis reveals significant changes of microbial compositions and protective functions during drinking water treatment. Scientific Reports, 3(1): 3550

Chapman J S (2003). Disinfectant resistance mechanisms, cross-resistance, and co-resistance. International Biodeterioration & Biodegradation, 51(4): 271–276

Chen S, Li X, Wang Y, Zeng J, Ye C, Li X, Guo L, Zhang S, Yu X (2018). Induction of *Escherichia coli* into a VBNC state through chlorination/chloramination and differences in characteristics of the bacterium between states. Water Research, 142: 279–288

Chiang W C, Nilsson M, Jensen P Ø, Høiby N, Nielsen T E, Givskov M, Tolker-Nielsen T (2013). Extracellular DNA shields against aminoglycosides in *Pseudomonas aeruginosa* biofilms. Antimicro-

bial Agents and Chemotherapy, 57(5): 2352–2361

Christgen B, Yang Y, Ahammad S Z, Li B, Rodriquez D C, Zhang T, Graham D W (2015). Metagenomics shows that low-energy anaerobic-aerobic treatment reactors reduce antibiotic resistance gene levels from domestic wastewater. Environmental Science & Technology, 49(4): 2577–2584

Conibear T C, Collins S L, Webb J S (2009). Role of mutation in *Pseudomonas aeruginosa* biofilm development. PLoS One, 4(7): e6289

D'Alessio M, Yoneyama B, Kirs M, Kisand V, Ray C (2015). Pharmaceutically active compounds: Their removal during slow sand filtration and their impact on slow sand filtration bacterial removal. Science of the Total Environment 524-525: 124–135

Dean R J, Shimmield T M, Black K D (2007). Copper, zinc and cadmium in marine cage fish farm sediments: an extensive survey. Environmental Pollution, 145(1): 84–95

Devi R, Alemayehu E, Singh V, Kumar A, Mengistie E (2008). Removal of fluoride, arsenic and coliform bacteria by modified homemade filter media from drinking water. Bioresource Technology, 99(7): 2269–2274

Driffield K, Miller K, Bostock J M, O'Neill A J, Chopra I (2008). Increased mutability of Pseudomonas aeruginosa in biofilms. J. Antimicrob. Chemother., 61(5): 1053–1056

Du M, Chen J, Sun F, Luan X, Wang D, Li Y, Zhang X (2008). Studies of viable but nonculturable Vibrio parahaemolyticus at low temperature under poor nutrition conditions and its resuscitation. Acta Hydrobiologica Sinica, 32(2): 178

Farkas A, Butiuc-Keul A, Ciatarâş D, Neamţu C, Crăciunaş C, Podar D, Drăgan-Bularda M (2013). Microbiological contamination and resistance genes in biofilms occurring during the drinking water treatment process. Science of the Total Environment, 443: 932–938

Friedberg E C, Walker G C, Siede W, Wood R D (2005). DNA repair and mutagenesis. Washington, DC: American Society for Microbiology Press

Garner E, Chen C, Xia K, Bowers J, Engelthaler D M, McLain J, Edwards M A, Pruden A (2018). Metagenomic characterization of antibiotic resistance genes in full-scale reclaimed water distribution systems and corresponding potable systems. Environmental Science & Technology, 52(11): 6113–6125

Geisenberger O, Ammendola A, Christensen B B, Molin S, Schleifer K H, Eberl L (1999). Monitoring the conjugal transfer of plasmid RP4 in activated sludge and *in situ* identification of the transconjugants. FEMS Microbiology Letters, 174(1): 9–17

Ghosh A, Singh A, Ramteke P W, Singh V P (2000). Characterization of large plasmids encoding resistance to toxic heavy metals in *Salmonella abortus equi*. Biochemical and Biophysical Research Communications, 272(1): 6–11

Gullberg E, Cao S, Berg O G, Ilbäck C, Sandegren L, Hughes D, Andersson D I (2011). Selection of resistant bacteria at very low antibiotic concentrations. PLoS Pathogens, 7(7): e1002158

Guo M T, Yuan Q B, Yang J (2015). Distinguishing effects of ultraviolet exposure and chlorination on the horizontal transfer of antibiotic resistance genes in municipal wastewater. Environmental Science & Technology, 49(9): 5771–5778

Guo X, Li J, Yang F, Yang J, Yin D (2014). Prevalence of sulfonamide and tetracycline resistance genes in drinking water treatment plants in the Yangtze River Delta, China. Science of the Total Environment, 493: 626–631

Han L, Liu W, Chen M, Zhang M, Liu S, Sun R, Fei X (2013). Comparison of NOM removal and microbial properties in up-flow/down-flow BAC filter. Water Research, 47(14): 4861–4868

Hasman H, Aarestrup F M (2002). tcrB, a gene conferring transferable copper resistance in *Enterococcus faecium*: Occurrence, transferability, and linkage to macrolide and glycopeptide resistance. Antimicrobial Agents and Chemotherapy, 46(5): 1410–1416

Hijnen W A M, Suylen G M H, Bahlman J A, Brouwer-Hanzens A, Medema G J (2010). GAC adsorption filters as barriers for viruses, bacteria and protozoan (oo)cysts in water treatment. Water Research, 44(4): 1224–1234

Hill D, Rose B, Pajkos A, Robinson M, Bye P, Bell S, Elkins M, Thompson B, Macleod C, Aaron S D, Harbour C (2005). Antibiotic susceptabilities of *Pseudomonas aeruginosa* isolates derived from patients with cystic fibrosis under aerobic, anaerobic, and biofilm conditions. Journal of Clinical Microbiology, 43(10): 5085–5090

Hoyle B D, Costerton J W (1991). Bacterial resistance to antibiotics: The role of biofilms. In: Progress in Drug Research. Boston: Birkhäuser Basel, 91–105

Hu Y, Jiang L, Zhang T, Jin L, Han Q, Zhang D, Lin K, Cui C (2018). Occurrence and removal of sulfonamide antibiotics and antibiotic resistance genes in conventional and advanced drinking water treatment processes. Journal of Hazardous Materials, 360: 364–372

Huang J J, Hu H Y, Tang F, Li Y, Lu S Q, Lu Y (2011). Inactivation and reactivation of antibiotic-resistant bacteria by chlorination in secondary effluents of a municipal wastewater treatment plant. Water Research, 45(9): 2775–2781

Icgen B, Yilmaz F (2014). Co-occurrence of antibiotic and heavy metal resistance in Kızılırmak River isolates. Bulletin of Environmental Contamination and Toxicology, 93(6): 735–743

Jia S, Shi P, Hu Q, Li B, Zhang T, Zhang X X (2015). Bacterial community shift drives antibiotic resistance promotion during drinking water chlorination. Environmental Science & Technology, 49(20): 12271–12279

Khan S, Beattie T K, Knapp C W (2016). Relationship between antibiotic- and disinfectant-resistance profiles in bacteria harvested from tap water. Chemosphere, 152: 132–141

Korotta-Gamage S M, Sathasivan A (2017). A review: Potential and challenges of biologically activated carbon to remove natural organic matter in drinking water purification process. Chemosphere, 167: 120–138

Legube B, Langlais B, Dore M (1981). Reactions of ozone with aromatics in dilute aqueous solution: Reactivity and biodegradability of oxidation products. In: Water Pollution Research and Development. Oxford: Pergamon Press, 553–570

Levin B R, Lipsitch M, Perrot V, Schrag S, Antia R, Simonsen L, Moore Walker N, Stewart F M (1997). The population genetics of antibiotic resistance. Clinical Infectious Diseases, 24(Supplement 1): S9–S16

Li C, Ling F, Zhang M, Liu W T, Li Y, Liu W (2017). Characterization of bacterial community dynamics in a full-scale drinking water treatment plant. Journal of Environmental Sciences (China), 51: 21–30

Li G, Ben W, Ye H, Zhang D, Qiang Z (2018). Performance of ozonation and biological activated carbon in eliminating sulfonamides and

sulfonamide-resistant bacteria: A pilot-scale study. Chemical Engineering Journal, 341: 327–334

Lin H, Ye C, Chen S, Zhang S, Yu X (2017). Viable but non-culturable *E. coli* induced by low level chlorination have higher persistence to antibiotics than their culturable counterparts. Environmental Pollution, 230: 242–249

Lin W, Zeng J, Wan K, Lv L, Guo L, Li X, Yu X (2018). Reduction of the fitness cost of antibiotic resistance caused by chromosomal mutations under poor nutrient conditions. Environment International, 120: 63–71

Liu A, Fong A, Becket E, Yuan J, Tamae C, Medrano L, Maiz M, Wahba C, Lee C, Lee K, Tran K P, Yang H, Hoffman R M, Salih A, Miller J H (2011). Selective advantage of resistant strains at trace levels of antibiotics: A simple and ultrasensitive color test for detection of antibiotics and genotoxic agents. Antimicrobial Agents and Chemotherapy, 55(3): 1204–1210

Liu J, Sun Q, Zhang C, Li H, Song W, Zhang N, Jia X (2016a). Removal of typical antibiotics in the advanced treatment process of productive drinking water. Desalination and Water Treatment, 57(24): 11386–11391

Liu S, Yang H, Liu W, Zhao Y, Wang X, Xie Y (2016b). Evaluation of backwash strategies on biologically active carbon filters by using chloroacetic acids as indicator chemicals. Process Biochemistry, 51(7): 886–894

Liu S S, Qu H M, Yang D, Hu H, Liu W L, Qiu Z G, Hou A M, Guo J, Li J W, Shen Z Q, Jin M (2018). Chlorine disinfection increases both intracellular and extracellular antibiotic resistance genes in a full-scale wastewater treatment plant. Water Research, 136: 131–136

Liu Y, Wang C, Tyrrell G, Hrudey S E, Li X F (2009). Induction of *Escherichia coli* O157:H7 into the viable but non-culturable state by chloraminated water and river water, and subsequent resuscitation. Environmental Microbiology Reports, 1(2): 155–161

Lu J, Tian Z, Yu J, Yang M, Zhang Y (2018). Distribution and abundance of antibiotic resistance genes in sand settling reservoirs and drinking water treatment plants across the Yellow River, China. Water (Basel), 10(3): 246

Ma X, Bibby K (2017). Free chlorine and monochloramine inactivation kinetics of *Aspergillus* and *Penicillium* in drinking water. Water Research, 120: 265–271

Madec J Y, Haenni M, Ponsin C, Kieffer N, Rion E, Gassilloud B (2016). Sequence type 48 *Escherichia coli* carrying the blaCTX-M-1 IncI1/ST3 plasmid in drinking water in France. Antimicrobial Agents and Chemotherapy, 60(10): 6430–6432

Madsen J S, Burmølle M, Hansen L H, Sørensen S J (2012). The interconnection between biofilm formation and horizontal gene transfer. FEMS Immunology and Medical Microbiology, 65(2): 183–195

Mah T F (2012). Biofilm-specific antibiotic resistance. Future Microbiology, 7(9): 1061–1072

Mao D, Luo Y, Mathieu J, Wang Q, Feng L, Mu Q, Feng C, Alvarez P J J (2014). Persistence of extracellular DNA in river sediment facilitates antibiotic resistance gene propagation. Environmental Science & Technology, 48(1): 71–78

McKinney C W, Pruden A (2012). Ultraviolet disinfection of antibiotic resistant bacteria and their antibiotic resistance genes in water and wastewater. Environmental Science & Technology, 46(24): 13393–13400

Molin S, Tolker-Nielsen T (2003). Gene transfer occurs with enhanced efficiency in biofilms and induces enhanced stabilisation of the biofilm structure. Current Opinion in Biotechnology, 14(3): 255–261

Murray G E, Tobin R S, Junkins B, Kushner D J (1984). Effect of chlorination on antibiotic resistance profiles of sewage-related bacteria. Applied and Environmental Microbiology, 48(1): 73–77

Narciso-da-Rocha C, Vaz-Moreira I, Svensson-Stadler L, Moore E R, Manaia C M (2013). Diversity and antibiotic resistance of *Acinetobacter* spp. in water from the source to the tap. Applied Microbiology and Biotechnology, 97(1): 329–340

Narkis N, Schneider-Rotel M (1980). Evaluation of ozone induced biodegradability of wastewater treatment plant effluent. Water Research, 14(8): 929–939

Negri M C, Lipsitch M, Blázquez J, Levin B R, Baquero F (2000). Concentration-dependent selection of small phenotypic differences in TEM-lactamase-mediated antibiotic resistance. Antimicrobial Agents and Chemotherapy, 44(9): 2485–2491

Nies D H (2000). Microbial heavy-metal resistance. Applied Microbiology and Biotechnology, 51: 451–460

Normander B, Christensen B B, Molin S, Kroer N (1998). Effect of bacterial distribution and activity on conjugal gene transfer on the phylloplane of the bush bean (Phaseolus vulgaris). Applied and Environmental Microbiology, 64(5): 1902–1909

Paulander W, Maisnier-Patin S, Andersson D I (2009). The fitness cost of streptomycin resistance depends on *rpsL* mutation, carbon source and RpoS (sigmaS). Genetics, 183(2): 539–546

Pinto A J, Xi C, Raskin L (2012). Bacterial community structure in the drinking water microbiome is governed by filtration processes. Environmental Science & Technology, 46(16): 8851–8859

Roberts A P, Pratten J, Wilson M, Mullany P (1999). Transfer of a conjugative transposon, Tn5397 in a model oral biofilm. FEMS Microbiology Letters, 177(1): 63–66

Rusin P, Gerba C (2001). Association of chlorination and UV irradiation to increasing antibiotic resistance in bacteria. Reviews of Environmental Contamination and Toxicology, 171: 1–52

Sakai A, Nakanishi M, Yoshiyama K, Maki H (2006). Impact of reactive oxygen species on spontaneous mutagenesis in *Escherichia coli.* Genes Cells, 11(7): 767–778

Seiler C, Berendonk T U (2012). Heavy metal driven co-selection of antibiotic resistance in soil and water bodies impacted by agriculture and aquaculture. Frontiers in Microbiology, 3: 399

Shi P, Jia S, Zhang X X, Zhang T, Cheng S, Li A (2013). Metagenomic insights into chlorination effects on microbial antibiotic resistance in drinking water. Water Research, 47(1): 111–120

Simazaki D, Kubota R, Suzuki T, Akiba M, Nishimura T, Kunikane S (2015). Occurrence of selected pharmaceuticals at drinking water purification plants in Japan and implications for human health. Water Research, 76: 187–200

Steinert M, Emödy L, Amann R, Hacker J (1997). Resuscitation of viable but nonculturable Legionella pneumophila Philadelphia JR32 by *Acanthamoeba castellanii*. Applied and Environmental Microbiology, 63(5): 2047–2053

Su H C, Liu Y S, Pan C G, Chen J, He L Y, Ying G G (2018). Persistence of antibiotic resistance genes and bacterial community changes in drinking water treatment system: From drinking water source to tap

water. Science of the Total Environment, 616-617: 453–461

Summers A O, Wireman J, Vimy M J, Lorscheider F L, Marshall B, Levy S B, Bennett S, Billard L (1993). Mercury released from dental "silver" fillings provokes an increase in mercury- and antibiotic-resistant bacteria in oral and intestinal floras of primates. Antimicrobial Agents and Chemotherapy, 37(4): 825–834

Tabak M, Scher K, Hartog E, Romling U, Matthews K R, Chikindas M L, Yaron S (2007). Effect of triclosan on *Salmonella typhimurium* at different growth stages and in biofilms. FEMS Microbiology Letters, 267(2): 200–206

Tawfik El-Zanfaly H, Reasoner D J, Geldreich E E (1998). Bacteriological changes associated with granular activated carbon in a pilot water treatment plant. Water, Air, and Soil Pollution, 107(1/4): 73–80

Turetgen I (2008). Induction of Viable but Nonculturable (VBNC) state and the effect of multiple subculturing on the survival of *Legionella pneumophila* strains in the presence of monochloramine. Annals of Microbiology, 58(1): 153–156

Van Acker H, Van Dijck P, Coenye T (2014). Molecular mechanisms of antimicrobial tolerance and resistance in bacterial and fungal biofilms. Trends in Microbiology, 22(6): 326–333

Walsh T R, Weeks J, Livermore D M, Toleman M A (2011). Dissemination of NDM-1 positive bacteria in the New Delhi environment and its implications for human health: An environmental point prevalence study. Lancet Infect Di, 11(5): 355–362

Walters M C 3rd, Roe F, Bugnicourt A, Franklin M J, Stewart P S (2003). Contributions of antibiotic penetration, oxygen limitation, and low metabolic activity to tolerance of *Pseudomonas aeruginosa* biofilms to ciprofloxacin and tobramycin. Antimicrobial Agents and Chemotherapy, 47(1): 317–323

Wan K, Zhang M, Ye C, Lin W, Guo L, Chen S, Yu X (2019). Organic carbon: An overlooked factor that determines the antibiotic resistome in drinking water sand filter biofilm. Environment International, 125: 117–124

Wang F, Li W, Li Y, Zhang J, Chen J, Zhang W, Wu X (2018). Molecular analysis of bacterial community in the tap water with different water ages of a drinking water distribution system. Frontiers of Environmental Science & Engineering, 12(3): 6

Weber W Jr, Pirbazari M, Melson G (1978). Biological growth on activated carbon: an investigation by scanning electron microscopy. Environmental Science & Technology, 12(7): 817–819

Wegrzyn G, Wegrzyn A (2002). Stress responses and replication of plasmids in bacterial cells. Microbial Cell Factories, 1(1): 2

Xi C, Zhang Y, Marrs C F, Ye W, Simon C, Foxman B, Nriagu J (2009). Prevalence of antibiotic resistance in drinking water treatment and distribution systems. Applied and Environmental Microbiology, 75 (17): 5714–5718

Xu L, Ouyang W, Qian Y, Su C, Su J, Chen H (2016). High-throughput profiling of antibiotic resistance genes in drinking water treatment plants and distribution systems. Environmental Pollution, 213: 119–126

Yang Y, Song W, Lin H, Wang W, Du L, Xing W (2018). Antibiotics and antibiotic resistance genes in global lakes: A review and meta-analysis. Environment International, 116: 60–73

Ye Z, Weinberg H S, Meyer M T (2007). Trace analysis of trimethoprim and sulfonamide, macrolide, quinolone, and tetracycline antibiotics in chlorinated drinking water using liquid chromatography electrospray tandem mass spectrometry. Analytical Chemistry, 79(3): 1135–1144

Yoon Y, Chung H J, Wen Di D Y, Dodd M C, Hur H G, Lee Y (2017). Inactivation efficiency of plasmid-encoded antibiotic resistance genes during water treatment with chlorine, UV, and UV/H_2O_2. Water Research, 123: 783–793

Zhang D, Li W, Zhang S, Liu M, Zhao X, Zhang X (2011). Bacterial community and function of biological activated carbon filter in drinking water treatment. Biomed. Environ. Sci., 24(2): 122–131

Zhang J, Li W, Chen J, Qi W, Wang F, Zhou Y (2018a). Impact of biofilm formation and detachment on the transmission of bacterial antibiotic resistance in drinking water distribution systems. Chemosphere, 203: 368–380

Zhang M, Chen L, Ye C, Yu X (2018b). Co-selection of antibiotic resistance via copper shock loading on bacteria from a drinking water bio-filter. Environmental Pollution, 233: 132–141

Zhang S, Ye C, Lin H, Lv L, Yu X (2015). UV disinfection induces a VBNC state in *Escherichia coli* and *Pseudomonas aeruginosa*. Environmental Science & Technology, 49(3): 1721–1728

Zhang S, Lin W, Yu X (2016). Effects of full-scale advanced water treatment on antibiotic resistance genes in the Yangtze Delta area in China. FEMS Microbiology Ecology, 92(5), fiw065

Zhang Y, Li A, Dai T, Li F, Xie H, Chen L, Wen D (2018c). Cell-free DNA: A neglected source for antibiotic resistance genes spreading from WWTPs. Environmental Science & Technology, 52(1): 248–257

Zheng J, Chen T, Chen H (2018). Antibiotic resistome promotion in drinking water during biological activated carbon treatment: Is it influenced by quorum sensing? Science of the Total Environment, 612: 1–8

Zhu X, Ye C, Wang Y, Chen L, Feng L (2019). Assessment of antibiotic resistance genes in dialysis water treatment processes. Frontiers of Environmental Science & Engineering, 13(3): 45